	Requester's Full Name: Khatol Sh	hmn-shah I	Examiner #: 78526 Date	8/28/51					
	Art Unit: 645 Phone Number 308 - 8896 Serial Number: 09/846 488 Mail Box and Bldg/Room Location: 80 - 17 Results Format Preferred (circle): PAPER DISK E-MAIL								
	3 ∈ -/2 If more than ne s arch is submit ***********************************	t d, pl as prioritize	search s in rder of ne d.	*****					
	************************************* Please provide a detailed statement of the se Include the elected species or structures, key utility of the invention. Define any terms the known. Please attach a copy of the cover shows the cover sha	earch topic, and describe as ywords, synonyms, acronyn at may have a special mear	specifically as possible the subject mans, and registry numbers, and combined in Give examples or relevant citations.	tter to be searched. with the concept or					
	Title of Invention:								
	Inventors (please provide full names):) See o	Hacked Bib	Sheet					
	Earliest Priority Filing Date:	4/25/2000		•					
•	*For Sequence Searches Only* Please include appropriate serial number.	all pertinent information (po	rent, child, divisional, or issued patent n	umbers) along with the					
		search	claims	6.1870					
Ĺ	is a k	1-29 Hacked	s claims and c	abstracts					
	nerozare nemena a parcocystis nemena a protestoria hughesi Fill neuspora hughesi ard protestocidal ard	Sourch	Including (eushors					
۸,	neuspoia humidal and	. meeting	p (ceit)	Jorena Pranmed Kulls dar hy Chavine Kulls avæn monkey Kulls monocyte					
	DUA (MANUMICA) -	Thanks Point of Contact:	hatil maider	darn Chainestille					
ENY	Te CM	Mary Hale echnical Info. Speciali 11 12D16 Tel: 308-42	st Canine Mous	monocy!					
egy	Merozoalal! Immunion Te CM Merozoalal! Immunion Te CM Merozoalal! Immunion Te CM	furn the Sear	attached po	hesus montey hide ne kidney by hamsler ledney					
	STAFF USE ONLY	Type of Search	Vendors and cost where a	pplicable					
	Searcher: 11 (11)	NA Sequence (#)	-STN 106. (C)						
	Searcher Phone #:	AA Sequence (#)	Questel/Orbit						
	Date Searcher Picked Up:	Bibliographic	Dr.Link	·					
	Date Completed:	Litigation	Lexis/Nexis						

Fulltext

Searcher Prep & Review Time:

Sequence Systems _

K. 5-Shah 840485

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)

SINCE FILE TOTAL ENTRY SESSION -7.06 -26.46

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FILE COVERS 1947 - 30 Aug 2001 VOL 135 ISS 10 FILE LAST UPDATED: 29 Aug 2001 (20010829/ED)

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This file supports REG1stRY for direct browsing and searching of all substance data from the REGISTRY file. Enter HELP FIRST for more information.

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E#	FREQUENCY	AТ	TERM				
E1	0	1	MEROTERPENOID/CT				
E2	0	3	MEROTERPENOID QUINONES/CT				
E3	0	2>	MEROZOITE/CT				
E4	0	2	MEROZOITE SURFACE PROTEIN 1/CT				
E5	. 0	1	MERPIQUAT/CT				
=> e e3+all/ct							
E1	0	> Me	erozoite/CT				
E2		USE	Development, microbial (L) merozoite/CT				
****	**** END*	**	-				

=> e sarcocystis neurona/ct 5

Page 60

Prepared by M. Hale 308-4258

```
E#
     FREQUENCY
                  AT
                          TERM
_--
                   _--
                          ____
                    5
                          SARCOCYSTIS MUCOSA/CT
E1
             1
                    5
                          SARCOCYSTIS MURIS/CT
E2
            18
                    5 --> SARCOCYSTIS NEURONA/CT
E3
            31
                          SARCOCYSTIS OVICANIS/CT
E4
             1
                    5
E5
             3
                          SARCOCYSTIS OVIFELIS/CT
=> e e3+all/ct
Ε1
          2314
                  BT4 Eukaryote (Eukaryotae)/CT
            75
                    BT3 Apicomplexa/CT
Ε2
            74
E3
                      BT2 Coccidia/CT
E4
            46
                        BT1 Sarcocystis/CT
E5
            31
                          -->
                               Sarcocystis neurona/CT
                            HN
                                  Valid heading during volume 116 (1992) to
                                  present.
*****
           END***
=> s e4-5
            46 SARCOCYSTIS/CT
            31 "SARCOCYSTIS NEURONA"/CT
L1
            71 (SARCOCYSTIS/CT OR "SARCOCYSTIS NEURONA"/CT)
=> e tachyzoite/ct 5
     FREQUENCY
                          TERM
Ε#
                   ΑT
__
     _____
                   __
                          ____
E1
             3
                   11
                          TACHYSURUS THALASSINUS/CT
E2
             2
                   11
                          TACHYSURUS ZONA/CT
              0
                      --> TACHYZOITE/CT
Ε3
                          TACITUS BELLUS/CT
E4
             1
                    1
                          TACKACE/CT
E5
             0
=> e neospora hughesi/ct
     FREQUENCY
                          TERM
E#
__
                    6
                          NEOSPORA/CT
E1
            30
E2
            97
                    5
                          NEOSPORA CANINUM/CT
E3
             6
                    5 --> NEOSPORA HUGHESI/CT
                          NEOSTENANTHERA/CT
Ε4
             n
                    8
E5
             1
                   8
                          NEOSTENANTHERA GABONENSIS/CT
             0
                   13
                          NEOSTIGMINE/CT
Ε6
             0
                    8
                          NEOSTREARIA/CT
E7
                    8
                          NEOSTREARIA FLECKERI/CT
E8
             1
           368
E9
                    2
                          NEOSTRIATUM/CT
                    2
E10
             0
                          NEOSTRIATUM (L) DOPAMINERGIC SYSTEM/CT
                    2
E11
              0
                          NEOSTRIATUM BRAIN/CT
                    2
              0
                          NEOSTRIATUM, DOPAMINERGIC SYSTEM BRAIN/CT
E12
=> e e3+all/ct
E1
                       Eukaryote (Eukaryotae)/CT
          2314
                  BT4
E2
            75
                    BT3 Apicomplexa/CT
E3
            74
                      BT2
                           Coccidia/CT
            30
E4
                        BT1 Neospora/CT
E5
             6
                          --> Neospora hughesi/CT
           END***
```

```
=> s e5
            6 "NEOSPORA HUGHESI"/CT
=> e protozocidal/ct 5
    FREQUENCY
               AΤ
                        TERM
__
                 --
                      PROTOZOAL INFECTION (L) SLEEPING SICKNESS/CT PROTOZOAN/CT
                 2
            0
E1
                 PROTUZUAM, C. --> PROTUZUAM, C. --> PROTUZUCIDAL/CT
E2
            0
E3
            0
E4
            0
                      PROTRACHEONISCUS/CT
                 12
                        PROTRACHEONISCUS ORIENTALIS/CT
E5
            2
=> e epm/ct 5
    FREQUENCY
                 AT
                      TERM
___
    ----
                 __
                        ----
                 2
E1
            0
                      EPK/CT
E2
            0
                 1
                      EPL/CT
E3
            0
                 1 --> EPM/CT
                 2
E4
            0
                      EPM 4050/CT
                 2
E5
            0
                       EPM 4060N/CT
=> e e3+all/ct
                --> EPM/CT
         0
****** END***
=> e equine protozoal myeloencephal?/ct 5
    FREQUENCY
               AT TERM
___
                 __
                 2 EQUINE MORBILLIVIRUS/CT
7 EQUINE PAPILLOMAVIRUS/CT
            0
E1
E2
            2
                  --> EQUINE PROTOZOAL MYELOENCEPHAL?/CT
E3
            0
E4
            2
                  6 EQUINE RHINOVIRUS/CT
                      EQUINE RHINOVIRUS 1/CT
E5
            1
                  6
=> e immune response/ct 5
    FREQUENCY AT
                      TERM
___
    _____
                 ___
                        ____
E1
            0
                 31
                      IMMUNE PROCESSES AND PHENOMENA (NON-CA HEADING)/CT
E2
            0
                 65
                      IMMUNE RECEPTORS (NON-CA HEADING)/CT
                 2 --> IMMUNE RESPONSE/CT
E3
            0
                 2
                     IMMUNE SERUMS/CT
E4
            0
                 2
                        IMMUNE SUPPRESSANTS/CT
E5
=> e e3+all/ct
                --> Immune response/CT
      0
                 USE Immunity/CT
       23219
***** END***
=> s e2
        23219 IMMUNITY/CT
=> e equine dermal/ct 5
  FREQUENCY AT
                        TERM
E#
    _____
                 __
```

E1		0	2		-		GONADOTROPIN/CT
E2		1				CORONAVIRU	S/CT
E3		0		>		DERMAL/CT	
E4		1	5		~	FOAMY VIRU	· ·
E5		0	2		EQUINE	HERPES 5 V	IRUS/CT
=> e	maiden	darby	bov	/ine	ct/		
em 21		1017	70 (17)		m D D N C		

E#	FREQUENCY	AT		TERM
E1	1	6	1	MAIASAURA PEEBLESORUM/CT
E2	0	1	1	MAICHUON/CT
E3	0		> 1	MAIDEN DARBY BOVINE/CT
E4	0	1	I	MAIDENHAIR/CT
E5	0	2	ĺ	MAIDENHAIR TREE/CT
E6	0	1	1	MAIDENI/CT
E7	0	1	I	MAIDENII/CT
E8	0	1	1	MAIDIDUS/CT
E9	0	1	1	MAIDIRADICIS/CT
E10	0	1	I	MAIDIS/CT
E11	0	2	1	MAIDISM/CT
E12	0	2	1	MAIDS/CT

=> fil medli

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	8.17	432.50
DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE	TOTAL
	ENTRY	SESSION
CA SUBSCRIBER PRICE	0.00	-26.46

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FILE LAST UPDATED: 28 AUG 2001 (20010828/UP). FILE COVERS 1958 TO DATE.

On April 22, 2001, MEDLINE was reloaded. See HELP RLOAD for details.

MEDLINE now contains IN-PROCESS records. See HELP CONTENT for details.

MEDLINE is now updated 4 times per week. A new current-awareness alert frequency (EVERYUPDATE) is available. See HELP UPDATE for more information.

MEDLINE thesauri in the /CN, /CT, and /MN fields incorporate the MeSH 2001 vocabulary. Enter HELP THESAURUS for details.

The OLDMEDLINE file segment now contains data from 1958 through 1965. Enter HELP CONTENT for details.

Left, right, and simultaneous left and right truncation are available in the Basic Index. See HELP SFIELDS for details.

THIS FILE CONTAINS CAS REGISTRY NUMBERS FOR EASY AND ACCURATE SUBSTANCE IDENTIFICATION.

=> e maiden darby bovine/ct

```
ΑT
                      TERM
E#
    FREQUENCY
--
    _____
                 __
                       ____
                     MAI INFECT/CT
           0
                 2
E1
                2
E2
            0
                      MAIC/CT
            0
                 --> MAIDEN DARBY BOVINE/CT
E3
            0
E4
                 1
                      MAIDENHAIR/CT
           0
                2
E5
                      MAIDENHAIR TREE/CT
           0
                2
                      MAIDS/CT
E6
           0
                2
                      MAIL/CT
E7
           0
                2
E8
                      MAIL DISTRIBUTION/CT
           0
                2
                      MAIL DISTRIBUTIONS/CT
E9
           0
                2
                      MAIL ORDER/CT
E10
                 2
E11
            Λ
                      MAIL, ELECTRONIC/CT
            0
                 2
                      MAIL-ORDER/CT
E12
=> e equine dermal/ct 5
    FREQUENCY
                       TERM
              AΤ
    _____
                 --
                       ____
--
                 2 EQUINE ARTERITIS VIRUSES/CT
2 EQUINE COITAL EXANTHEMA VIRUS/CT
           0
E1
                2
           0
E2
            0
E3
                 --> EQUINE DERMAL/CT
            0
                 2
                      EQUINE DISEASE/CT
£4
            0
                 2
                       EQUINE DISEASES/CT
E5
=> e canine monocyte/ct 5
E#
   FREQUENCY AT
                       TERM
___
    _____
                 2
           0
                       CANINE INFECTIOUS HEPATITIS/CT
E1
                     CANINE INFECTIOUS HEPATITIS VIRUS/CT
                 2
            0
E2
            0
                 --> CANINE MONOCYTE/CT
E3
            0
                 2
                      CANINE PARVOVIRUS/CT
E4
E5
            0
                 2
                       CANINE PARVOVIRUSES/CT
=> e mouse monocyte/ct 5
    FREQUENCY AT
                       TERM
                 ---
                       ----
___
E1
            0
                 2
                       MOUSE LYMPHOCYTE PROTEIN MOIETY REDUCED OF
INTERFERON
                       TYPE II/CT
            0
                 2
                      MOUSE MAMMARY TUMOR VIRUS/CT
E2
                  --> MOUSE MONOCYTE/CT
E3
            0
            0
                 2
                      MOUSE MUTANT STRAIN/CT
E4
            0 .
E5
                 2
                      MOUSE MUTANT STRAINS/CT
=> e fetal rhesus monkey kidney/ct 5
  FREQUENCY
              ΑT
E#
                       TERM
    -----
___
           40
                       FETAL RESORPTION: VE, VETERINARY/CT
E.1
                     FETAL RESORPTIONS/CT
E2
           n
                 2
                  --> FETAL RHESUS MONKEY KIDNEY/CT
E3
            n
                      FETAL STRUCTURE/CT
            0
                 2
F.4
E5
            0
                 2
                      FETAL STRUCTURES/CT
=> e feline kidney/ct 5
   FREQUENCY
E#
              AT
                       TERM
```

```
7
                        FELINE INFECTIOUS PERITONITIS: TM, TRANSMISSION/CT
E1
                       FELINE INFECTIOUS PERITONITIS: VI, VIROLOGY/CT
E2
           16
            0
                   --> FELINE KIDNEY/CT
E3
            0
                  2 FELINE LENTIVIRUS/CT
E4
                 2
E5
            0
                        FELINE LENTIVIRUSES/CT
=> fil medl, caplus, biosis, embase, wpids, jicst
                                                SINCE FILE
                                                                TOTAL
COST IN U.S. DOLLARS
                                                     ENTRY
                                                              SESSION
                                                              433.10
                                                      0.60
FULL ESTIMATED COST
                                                SINCE FILE
                                                               TOTAL
DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)
                                                     ENTRY SESSION
CA SUBSCRIBER PRICE
                                                      0.00
                                                              -26.46
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FILE 'EMBASE' ENTERED AT 11:01:09 ON 30 AUG 2001
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FILE 'WPIDS' ENTERED AT 11:01:09 ON 30 AUG 2001
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FILE 'JICST-EPLUS' ENTERED AT 11:01:09 ON 30 AUG 2001
COPYRIGHT (C) 2001 Japan Science and Technology Corporation (JST)
=> s (11 or 12 or merozoite? or sarcocystis neurona or tachyzoite or neospor?
hughes? or protozocid?)
          3707 FILE MEDLINE
L5
         1804 FILE CAPLUS
L6
          3473 FILE BIOSIS
L7
         2743 FILE EMBASE
          108 FILE WPIDS
rs
          179 FILE JICST-EPLUS
TOTAL FOR ALL FILES
        12014 (L1 OR L2 OR MEROZOITE? OR SARCOCYSTIS NEURONA OR TACHYZOITE
L10
OR
               NEOSPOR? HUGHES? OR PROTOZOCID?)
=> s (epm or equine protozoa? myeloencephal?)
L11
          376 FILE MEDLINE
L12
          856 FILE CAPLUS
L13
          472 FILE BIOSIS
L14
          332 FILE EMBASE
L15
          380 FILE WPIDS
```

```
L16
            75 FILE JICST-EPLUS
TOTAL FOR ALL FILES
L17
          2491 (EPM OR EQUINE PROTOZOA? MYELOENCEPHAL?)
=> s (equine derma? or maiden darby (w) (canine or bovine) (w) kidney or african
green monkey kidney or (canine or mouse) (w) monocyte or fetal rhesus monkey
kidney or feline kidney or baby hamster kidney) (2a) cell!
          1449 FILE MEDLINE
L19
          1645 FILE CAPLUS
L20
          6592 FILE BIOSIS
L21
          1282 FILE EMBASE
L22
            75 FILE WPIDS
L23
            39 FILE JICST-EPLUS
TOTAL FOR ALL FILES
         11082 (EQUINE DERMA? OR MAIDEN DARBY (W) (CANINE OR BOVINE) (W) KIDNEY
               OR AFRICAN GREEN MONKEY KIDNEY OR (CANINE OR MOUSE) (W)
MONOCYTE
               OR FETAL RHESUS MONKEY KIDNEY OR FELINE KIDNEY OR BABY HAMSTER
               KIDNEY) (2A) CELL!
=> s 110 and 117 and 124
             1 FILE MEDLINE
             O FILE CAPLUS
L26
L27
             1 FILE BIOSIS
L28
             1 FILE EMBASE
L29
             1 FILE WPIDS
L30
             O FILE JICST-EPLUS
TOTAL FOR ALL FILES
             4 L10 AND L17 AND L24
=> dup rem 131
PROCESSING COMPLETED FOR L31
              2 DUP REM L31 (2 DUPLICATES REMOVED)
=> d cbib abs 1-2
    ANSWER 1 OF 2 WPIDS COPYRIGHT 2001
                                            DERWENT INFORMATION LTD
ΑN
     2000-571969 [53]
                        WPIDS
CR
     2001-218486 [22]
AΒ
     WO-200049049 A UPAB: 20010421
     NOVELTY - Detection of Sarcocystis neurona in horses
     new.
```

by identifying a specific antibody-antigen complex via an immunoassay is

DETAILED DESCRIPTION - Detection of Sarcocystis neurona in an equine in an immunoassay is improved by reacting a biological sample from the horse suspected of harboring the S. neurona with an antibody (Ab) which is selective in binding to an identifying S. neurona antigen (Ag) to form an Ab-Ag complex.

- INDEPENDENT CLAIMS are also included for the following:
- (1) a kit for detecting S. neurona in a biological sample from an equine;
 - (2) monoclonal antibodies against 16 plus or minus 4 kDa or 30 plus

```
(3) isolated DNA sequences encoding the 16 plus or minus 4 kDa and
30
     plus or minus 4 kDa antigens of S. neurona.
          USE - The methods and antibodies are useful for detecting S. neurona
     (claimed) which causes equine protozoal
     myeloencephalitis, a neurological disorder in horses.
     Dwg.0/0
                                                        DUPLICATE 1
L32 ANSWER 2 OF 2
                       MEDLINE
2000043702 Document Number: 20043702.
                                        PubMed ID: 10577742.
                                                                Simplified
     technique for isolation, excystation, and culture of Sarcocystis species
     from opossums. Murphy A J; Mansfield L S. (Animal Health Diagnostic
     Laboratory, Michigan State University, East Lansing 48824, USA. ) JOURNAL
    OF PARASITOLOGY, (1999 Oct) 85 (5) 979-81. Journal code: JL3; 7803124.
     ISSN: 0022-3395. Pub. country: United States. Language: English.
     Sarcocystis neurona is a protozoan parasite that
AΒ
     causes a neurological disease in horses called equine
     protozoal myeloencephalitis. The route of transmission
     is speculated to be by fecal-oral transfer of sporocysts shed from
     opossums. Controversy exists regarding both the natural life cycle for
     this parasite as well as the species identity of opossum Sarcocystis. To
     provide stage-specific material for species comparison, 27 opossums from
     southern Michigan were screened for Sarcocystis spp. sporocysts. Seven
     opossums were positive for Sarcocystis sporocysts by fecal flotation. A
     simplified, effective technique for isolation, excystation, and culture
of
     opossum Sarcocystis sp. from mucosal scrapings was developed. All 7
     Sarcocystis sp. isolates were successfully cultured to grow long term in
     equine dermal cells to the merozoite
     stage. Merozoites were observed between 5 and 15 days after
     inoculation. In conclusion, opossums shed Sarcocystis sp. sporocysts that
     may be manipulated to excyst and grow in vitro in equine dermal cell
lines
     to the merozoite stage using the simplified technique described.
=> s 110 and 117 and (13 or immunity or immune response or immunogen?)
L33
             5 FILE MEDLINE
L34
             3 FILE CAPLUS
             3 FILE BIOSIS
L35
L36
             4 FILE EMBASE
L37
             1 FILE WPIDS
L38
             O FILE JICST-EPLUS
TOTAL FOR ALL FILES
            16 L10 AND L17 AND (L3 OR IMMUNITY OR IMMUNE RESPONSE OR
IMMUNOGEN?
=> s 139 not 131
L40
             5 FILE MEDLINE
             3 FILE CAPLUS
L41
L42
             3 FILE BIOSIS
L43
             4 FILE EMBASE
```

or minus 4 kDa antigens of S. neurona; and

```
1 FILE WPIDS
L44
L45
             O FILE JICST-EPLUS
TOTAL FOR ALL FILES
           16 L39 NOT L31
=> dup rem 146
PROCESSING COMPLETED FOR L46
              8 DUP REM L46 (8 DUPLICATES REMOVED)
L47
=> d cbib abs 1-8
L47 ANSWER 1 OF 8 CAPLUS COPYRIGHT 2001 ACS
             Document No. 134:221431 Vaccine to control equine
    protozoal myeloencephalitis in horses. Mansfield, Linda/
     S.; Rossano, Mary G.; Murphy, Alice J.; Vrable, Ruth A/ (Michigan State
     University, USA). PCT Int. Appl. WO 2001015708 A1 20010308, 57 pp.
     DESIGNATED STATES: W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA,
CH,
     CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN,
     IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN,
     MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT,
     UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM; RW: AT,
     BE, BF, BJ, CF, CG, CH, CI, CM, CY, DE, DK, ES, FI, FR, GA, GB, GR, IE,
     IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG.
                                                     (English). CODEN:
     PIXXD2. APPLICATION: WO 2000-US24221 20000831. PRIORITY: US
     1999-PV152193 19990902; US 2000-513086 20000224.
     The present invention provides vaccines and methods for making the
     vaccines that actively or passively protect an equid or other animal
     against Sarcocystis neurona. In particular, the
     present invention provides vaccines that provide active immunity
     which comprise a polypeptide or DNA vaccine that contains or expresses at
     least one epitope of an antigen that has an amino acid sequence
     substantially similar to a unique 16 (+/-4) kDa antigen and/or 30 (+/-4)
     kDa antigen of Sarcocystis neurona. The present
     invention further provides a vaccine that provides passive
     immunity to Sarcocystis neurona comprising
     polyclonal or monoclonal antibodies against at least one epitope of an
     antigen substantially similar to a unique 16 (+/-4) kDa antigen and/or 30
     (+/-4) kDa antigen of Sarcocystis neurona.
L47 ANSWER 2 OF 8
                       MEDLINE
                                                        DUPLICATE 1
```

L47 ANSWER 2 OF 8 MEDLINE DUPLICATE 1
2001434376 Document Number: 21125368. PubMed ID: 11226451. Molecular comparison of the dense granule proteins GRA6 and GRA7 of Neospora hughesi and Neospora caninum. Walsh C P; Vemulapalli R; Sriranganathan N; Zajac A M; Jenkins M C; Lindsay D S. (Center for Molecular Medicine and Infectious Diseases, Department of Biomedical Sciences and Pathobiology, Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, VA 24061-0342, USA.) INTERNATIONAL JOURNAL FOR PARASITOLOGY, (2001 Mar) 31 (3) 253-8. Journal code: GSB; 0314024. ISSN: 0020-7519. Pub. country: England: United Kingdom.

English.

AB Neospora hughesi is a recently described apicomplexan parasite that has been associated with several cases of equine

protozoal myeloencephalitis. The biology of this new
parasite is just beginning to be defined. Towards this understanding, we
report important differences between the nucleotide and deduced amino

acid

sequences of the dense granule proteins GRA6 and GRA7 of N. hughesi and Neospora caninum. This information can be used to differentiate the two species and contribute to further understanding of the prevalence and biology of N. hughesi. The newly defined proteins of N. hughesi are referred to as NhGRA6 and NhGRA7 in keeping with the protocol for naming homologous proteins of the Apicomplexa. Genes of the two dense granule proteins of N. hughesi (isolate Nh-Al) and four different isolates of N. caninum were isolated via PCR and their DNA sequences were determined. Computer analysis indicated that the two gene sequences were identical among all four N. caninum isolates. However, the gene for NhGRA6 was

found

of

to be 96 nucleotides longer at the 3' end than that of NcGRA6, resulting in a protein product that is 32 amino acids larger than NcGRA6. Two tandem

repeat sequences were identified at the 3' end of the NhGRA6 gene. These repeat sequences contributed to the lengthening of the carboxy terminus

NhGRA6 in comparison with that of NcGRA6. The larger size of NhGRA6 was further confirmed by Western blot analysis in which NcGRA6 monospecific antibodies recognised a protein of approximately 42 kDa in N. hughesi whole tachyzoite preparation but a protein of 37 kDa in N. caninum whole tachyzoite preparation. Analysis of GRA7 gene sequences indicated a 6 and 14.8% difference at nucleotide and amino acid sequence level, respectively, between NcGRA7 and NhGRA7. Despite the same number of residues in the deduced amino acid sequences of all the GRA7 proteins, Western blot analysis indicated a difference in the migration pattern of NhGRA7 in comparison with NcGRA7. Results of our study

indicate

that diagnostic tests based on differences in dense granule sequences and antigenicity may have potential to differentiate between N. hughesi and N.

caninum. Such diagnostic tests would be valuable tools to aid in our understanding of the epidemiology of these parasites. Additionally, dense granule proteins are immunogenic and they may have potential as use in recombinant vaccines against neosporosis.

L47 ANSWER 3 OF 8 MEDLINE DUPLICATE 2
2001354018 Document Number: 21127318. PubMed ID: 11223200.

Immunoconversion against Sarcocystis neurona in normal and dexamethasone-treated horses challenged with S. neurona sporocysts.

Cutler T J; MacKay R J; Ginn P E; Gillis K; Tanhauser S M; LeRay E V; Dame

J B; Greiner E C. (Department of Pathobiology, PO Box 100880, College of Veterinary Medicine, University of Florida, Gainesville 32610, USA.) VETERINARY PARASITOLOGY, (2001 Feb 26) 95 (2-4) 197-210. Journal code: XBU; 7602745. ISSN: 0304-4017. Pub. country: Netherlands. Language: English.

AB Equine protozoal myeloencephalitis is a common neurologic disease of horses in the Americas usually caused by Sarcocystis neurona. To date, the disease has not been induced in horses using characterized sporocysts from Didelphis

virginiana, the definitive host. S. neurona sporocysts from 15 naturally infected opossums were fed to horses seronegative for antibodies against S. neurona. Eight horses were given 5x10(5) sporocysts daily for 7 days. Horses were examined for abnormal clinical signs, and blood and cerebrospinal fluid were harvested at intervals for 90 days after the first day of challenge and analyzed both qualitatively (western blot) and quantitatively (anti-17kDa) for anti-S. neurona IgG. Four of the challenged horses were given dexamethasone (0.1mg/kg orally once daily) for the duration of the experiment. All challenged horses immunoconverted against S. neurona in blood within 32 days of challenge and in CSF within 61 days. There was a trend (P = 0.057) for horses given dexamethasone to immunoconvert earlier than horses that were not immunosuppressed. Anti-17kDa was detected in the CSF of all challenged horses by day 61. This response was statistically greater at day 32 in horses given dexamethasone. Control horses remained seronegative throughout the period in which all challenged horses converted. One control horse immunoconverted in blood at day 75 and in CSF at day 89. Signs of neurologic disease were mild to equivocal in challenged horses. Horses given dexamethasone had more severe signs of limb weakness than did

horses

not given dexamethasone; however, we could not determine whether these signs were due to spinal cord disease or to effects of systemic illness. At necropsy, mild-moderate multifocal gliosis and neurophagia were found histologically in the spinal cords of 7/8 challenged horses. No organisms were seen either in routinely processed sections or by immunohistochemistry. Although neurologic disease comparable to naturally occurring equine protozoal myeloencephalitis (EPM) was not produced, we had clear evidence of an immune response to challenge both systemically and in the CNS. Broad immunosuppression with dexamethasone did not increase the severity of histologic changes in the CNS of challenged horses. Future work must focus on defining the factors that govern progression of inapparent S. neurona infection to EPM.

L47 ANSWER 4 OF 8 MEDLINE 2001354017 Document Number: 21127317. PubMed ID: 11223199. Interpretation

of the detection of Sarcocystis neurona antibodies in the serum of young horses. Cook A G; Buechner-Maxwell V; Morrow J K; Ward D L; Parker N A; Dascanio J J; Ley W B; Cooper W. (Department of Large Animal Clinical Sciences, Virginia-Maryland College of Veterinary Medicine, Duck Pond Drive Phase II, Blacksburg, VA 24061, USA.. ancook2@vt.edu) . VETERINARY PARASITOLOGY, (2001 Feb 26) 95 (2-4) 187-95. Journal code: XBU; 7602745. ISSN: 0304-4017. Pub. country: Netherlands. Language: English.

AB Horses that are exposed to Sarcocystis neurona, a causative agent of equine protozoal myeloencephalitis, produce antibodies that are detectable in serum by western blot (WB). A positive test is indicative of exposure to the organism. Positive tests in young horses can be complicated by the presence of maternal antibodies. Passive transfer of maternal antibodies to S. neurona from seropositive mares to their foals was evaluated. Foals were sampled at birth (presuckle), at 24h of age (postsuckle), and at monthly intervals. All foals sampled before suckling were seronegative. Thirty-three foals from 33 seropositive mares became seropositive with

colostrum ingestion at 24h of age, confirming that passive transfer of S. neurona maternal antibodies occurs. Thirty-one of the 33 foals became seronegative by 9 months of age, with a mean seronegative conversion time of 4.2 months. These results indicate that evaluation of exposure to S. neurona by WB analysis of serum may be misleading in young horses.

L47 ANSWER 5 OF 8 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V. 2001075066 EMBASE A review of Sarcocystis neurona and

equine protozoal myeloencephalitis (

EPM). Dubey J.P.; Lindsay D.S.; Saville W.J.A.; Reed S.M.; Granstrom D.E.; Speer C.A.. J.P. Dubey, United States Dept. of Agriculture, Animal and Natural Resources Inst., Beltsville Agricultural Res. Center, Beltsville, MD 20705-2350, United States. jdubey@anri.barc.usda.gov. Veterinary Parasitology 95/2-4 (89-131) 26 Feb 2001.

Refs: 148.

ISSN: 0304-4017. CODEN: VPARDI.

Publisher Ident.: S 0304-4017(00)00384-8. Pub. Country: Netherlands.

Language: English. Summary Language: English.

AΒ Equine protozoal myeloencephalitis (EPM) is a serious neurological disease of horses in the Americas. The protozoan most commonly associated with EPM is

Sarcocystis neurona. The complete life cycle of S. neurona is unknown, including its natural intermediate host that harbors its sarcocyst. Opossums (Didelphis virginiana, Didelphis albiventris) are its definitive hosts. Horses are considered its aberrant hosts because only schizonts and merozoites (no sarcocysts) are found in horses. EPM-like disease occurs in a variety of mammals including cats, mink, raccoons, skunks, Pacific harbor seals, ponies, and Southern sea otters. Cats can act as an experimental intermediate host harboring the sarcocyst stage after ingesting sporocysts. This paper reviews information on the history, structure, life cycle, biology, pathogenesis, induction of disease in animals, clinical signs, diagnosis, pathology, epidemiology, and treatment of EPM caused by S. neurona.

L47 ANSWER 6 OF 8 MEDLINE

2001156688 Document Number: 21088454. PubMed ID: 11219340. protozoal myeloencephalitis. MacKay R J; Granstrom D E; Saville W J; Reed S M. (Department of Large Animal Clinical Sciences, College of Veterinary Medicine, University of Florida, Gainesville, Florida, USA.) VETERINARY CLINICS OF NORTH AMERICA. EQUINE PRACTICE, (2000 Dec) 16 (3) 405-25. Ref: 96. Journal code: CEP; 8511904. ISSN: 0749-0739. Pub. country: United States. Language: English.

AΒ Recent advances in the understanding of the parasite life cycle, epidemiology, clinical signs, diagnosis, treatment, and prevention of EPM are reviewed. The NAHMS Equine '98 study and a controlled retrospective study from The Ohio State University College of Veterinary Medicine identified a number of risk factors associated with development of the disease. The national annual incidence of EPM was 1% or less depending on the primary use of the animals. Increased disease risk was associated with age (1-5 and > 13 years of age), season (lowest in winter months and increasing with ambient temperature), previous

events, the presence of opossums, the use of nonsurface water drinking

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systems, and failure to restrict wildlife access to feed. Horses that received treatment were 10 times more likely to improve, and those that improved were 50 times more likely to survive. A number of recent studies confirmed that horses can be experimentally infected with S. neurona; however, large numbers of sporocysts are apparently necessary to achieve infection, and clinical signs and abnormal CNS histology are only seen inconsistently. Results suggest that CNS infection and positive CSF immunoblot findings may be transient phenomena among naturally infected horses. Although immunosuppression may be involved in the development of EPM, some element of the immune response seems

to be necessary for the development of clinical signs. Use of the standard $% \left(1\right) =\left(1\right) +\left(1\right)$

immunoblot test for the detection of anti-S. neurona antibodies in CSF continues to provide the most useful adjunct to a detailed neurologic examination for the diagnosis of EPM. Test sensitivity and specificity were 89% in 295 horses euthanatized because of neurologic disease, of which 123 were confirmed cases of EPM. The PPV was 85%, and the NVP was 92%. A number of promising new EPM treatments are under investigation. In addition to standard SDZ/PYR therapy, toltrazuril, ponazuril, diclazuril, and NTZ have shown promise

as

possible alternatives.

L47 ANSWER 7 OF 8 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD

AN 1999-571872 [48] / WPIDS

NOVELTY - Biologically pure culture of equine Neospora, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the

following: (a) detecting antibodies (Ab) specifically reactive with equine

- Neospora antigens (Ag) by forming an Ab-Ag complex;

 (b) detecting Neospora by forming a complex with an antibody (Ab1)
- specifically reactive with Neospora antigen;

 (c) detecting Neospora-specific nucleic acid (I) by hybridization
- with a specific oligonucleotide probe; and

 (d) pharmaceutical composition containing equipe Neospora
- (d) pharmaceutical composition containing equine Neospora immunogen and a carrier.

ACTIVITY - Antiprotozoal.

 ${\tt MECHANISM}$ OF ACTION - Induction of a specific ${\tt immune}$ ${\tt response}.$

USE - Immunogens (optionally expressed from gene therapy vectors) from equine Neospora are used in vaccines for treatment or prevention of Neospora infection in horses and other animals. Neospora is a causative agent of equine protozoal

myeloencephalitis (EPM). Detection of Neospora-specific antigens, antibodies or nucleic acid (by usual immunoassay or hybridization tests) is used to diagnose infection. Antibodies (Ab) specific for equine Neospora are used for diagnosis; to select candidate immunogens for vaccine development; to isolate proteins; to screen DNA libraries and as therapeutic/prophylactic agents.

ADVANTAGE - Reagents specific for equine Neospora allow differentiation between equine protozoal myeloencephalitis caused by Neospora and Sarcocystis neurona. These pathogens require different treatments and treatment of Neospora is only effective if applied before the parasite

has

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formed cysts. The vaccines also prevent shedding of oocysts by animals known to be infected. $\ensuremath{\text{Dwg.0/2}}$

L47 ANSWER 8 OF 8 MEDLINE DUPLICATE 3
1998234002 Document Number: 98234002. PubMed ID: 9573058. Evidence that surface proteins Sn14 and Sn16 of Sarcocystis neurona merozoites are involved in infection and immunity. Liang F T; Granstrom D E; Zhao X M; Timoney J F. (Gluck Equine Research Center, Department of Veterinary Science, University of Kentucky, Lexington 40546-0099, USA.) INFECTION AND IMMUNITY, (1998 May) 66 (5) 1834-8. Journal code: GO7; 0246127. ISSN: 0019-9567. Pub. country: United States. Language: English.

AB Sarcocystis neurona is the etiologic agent of
equine protozoal myeloencephalitis (
EPM). Based on an analysis of 25,000 equine serum and
cerebrospinal fluid (CSF) samples, including samples from horses with
neurologic signs typical of EPM or with histologically or
parasitologically confirmed EPM, four major immunoblot band
patterns have been identified. Twenty-three serum and CSF samples
representing each of the four immunoblot patterns were selected from 220
samples from horses with neurologic signs resembling EPM and

samples from horses with neurologic signs resembling EPM and examined for inhibitory effects on the infectivity of S. neurona by an in vitro neutralization assay. A high correlation between immunoblot band pattern and neutralizing activity was detected. Two proteins, Sn14 and Sn16 (14 and 16 kDa, respectively), appeared to be important for in vitro infection. A combination of the results of surface protein labeling, immunoprecipitation, Western blotting, and trypsin digestion suggests

that

these molecules are surface proteins and may be useful components of a vaccine against S. neurona infection. Although S. neurona is an obligate intracellular parasite, it is potentially a target for specific antibodies

which may lyse merozoites via complement or inhibit their attachment and penetration to host cells.

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L81
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